WEST

End of Result Set

Generate Collection Print

L16: Entry 4 of 14 File: DWPI

DERWENT-ACC-NO: 2000-XXXXXX

DERWENT-WEEK: 200014

COPYRIGHT 2006 DERWENT INFORMATION LTD

TITLE: Method and particular [illegible] of polynucleotides having a diagnostic/therapeutic protein A activity, useful for studying the structure, activity relationship of the protein, and for identifying [illegible] drugs

INVENTOR: MIMAKI, Y; OHASHIMA, K

PATENT-ASSIGNEE: ASKA RSRC LAB KK [ASKAN]; ASKA SEIYAKU KK [ASKAN]

PRIORITY-DATA: 1999JP-0219448 (August 17, 1999)

PATENT FAMILY:

PUB-NO	PUB-DATE	LANGUAGE	PAGES	MAIN-IPC
DE 69901274 E	October 1, 2002		110	C12N015-74
AU 2000019716 A1	February 14, 2001	E	041	C12N015-74
AU 746961 A	March 6, 2002		000	C12N015-74
EP 1106608 A1	June 13, 2001	E	000	C12N015-74
US 20020061513 A1	May 23, 2002		000	C12Q001-00
EP 1231276 A1	August 14, 2002	E	000	C12N015-74
JP 2003526308 W	July 23, 2003		039	C12N015-09
EP 1106608 B1	September 4, 2002	E	000	C12N015-74

DESIGNATED-STATES: AR AU AM AT AT AU CA CH CN CR CU CZ DE DK DM DZ EC EE ES FI GB GD GE GH GM HR HU ID IL IN IS JP KE KG KP KR KZ LC LK LR LS LT LU LV MA MD MG MK MN MW MX NO NZ PL PT RO RU SD SE SG SI SK SL TJ TM TR TT TZ UA UG US UZ VN YU ZA ZW AT BE CH CY DE DK EA ES FI FR GB GR IE IT LU MC NL PT SE TR AT BE CH CY DE DK EA ES FI FR GB GR IE IT LU MC NL PT SE AT BE CH CY DE DK EA ES FI FR GB GR IE IT LI LU MC NL PT SE

APPLICATION-DATA:

PUB NO	APPL DATE	APPL NO	RELATIONSHIP
DE 6[illegible]E	August 17, 1999	1999EP [illegible]	
DE 6[illegible]E	August 17, 1999	1999EP 0944[illegible]	
DE 6[illegible]E	August 17, 1999	1999WO EP[illegible]	
DE 6[illegible]E		EP 11[illegible]	Based on
DE 6[illegible]E		WO [illegible]	Based on
AU [illegible]A	August 17, 1999	1999WO EP[illegible]	
AU [illegible]A	August 17, 1999	1999AU [illegible]	
AU [illegible]A		WO [illegible]	Based on
EP 1105508A1	August 17, 1999	1999EP 0944420	
EP 1105508A1	August 17, 1999	1999WO EP06022	
EP 1105508A1		WO 200009716	Based on
US2002006151[illegible]A1	August 17, 1999	1999WO EP06022	Cont of
US2002006151[illegible]A1	February 14, 2001	2001US 0785[illegible]	
EP 1231274A1	August 17, 1999	1999EP 0944420	Div ex
EP 1231274A1	August 17, 1999	2000EP 0004765	
EP 1231274A1		EP 1105508	Div ex
JP2002522058W	August 17, 1999	1999WO EP06022	
JP2002522058W	August 17, 1999	2000JP 0565[illegible]	
JP2002522058W		WO 200009716	Based on
EP 1105508B1	August 17, 1999	1999EP 0944420	
EP 1105508B1	August 17, 1999	1999WO EP06022	
EP 1105508B1	August 17, 1999	2000EP 0004765	Related to
EP 1105508B1		EP 1231274	Related to
EP 1105508B1		WO 200009716	Based on

INT CL (IPC): [illegible]; C12N 1/19; C12N 1/21; C12N 5/10; C12N 15/09; C12N 15/11; C12N 15/62; C12N 15/74; C12P 21/02; C12P 21/02; C12P 21/04; C12Q 1/[illegible]; C12Q 1/02; C12Q 1/37; C12Q 1/68; G01N 33/53; G01N 33/566

ABSTRACTED-PUB-NO: EP 1105508B
BASIC-ABSTRACT:

NOVELTY - A method for detecting or purifying substances uses polypeptides or subunits fused to at least 2 different affinity tags, at least one of which is an IgG binding domain of Staphylococcus protein A (SPA).

DETAILED DESCRIPTION - A method of detecting and/or purifying substances selected from proteins, [illegible], complexes of proteins or [illegible] subunits, cell components, cell organelles and cells, comprises:

(a) providing an expression environment containing one or more heterologous nucleic acids encoding polypeptides and/or subunits of a protein complex, the polypeptides or subunits being fused to at least 2 different affinity tags, one of which consists of one or more IgG binding domains of SPA;

(b) maintaining the expression environment to express the polypeptides or subunits in a native form as fusion proteins with the affinity tags; and

(c) detecting and/or purifying the polypeptide or subunits by a combination of at least 2 affinity purification steps, each comprising binding the polypeptides or subunits via the affinity tag to a support material capable of selectively binding one of the affinity tags and separating the polypeptides or subunits from the support material after the substances have been removed.

INDEPENDENT CLAIMS are also included for the following:

(1) [illegible]

Claims

1. A process for purifying a protein complex comprising at least two different affinity tags, which process comprises applying a solution containing the protein complex to a support material capable of selectively binding one of the affinity tags and separating the complex from the support material after unbound substances have been removed;

2. A fusion protein comprising at least one polypeptide or subunit of a protein complex fused to at least 2 different affinity tags, where one of the affinity tags consists of at least two IgG binding domains of SPA;

3. A nucleic acid coding for a fusion protein of claim 2;

4. A vector comprising a nucleic acid as in claim 3 under the control of sequences facilitating the expression of a fusion protein as in claim 2;

5. A vector comprising heterologous nucleic acid sequences in form of one or more cassettes each comprising at least 2 different affinity tags, one consisting of one or more IgG binding domains of Staphylococcus aureus protein A (SPA), and at least one MCS linker for the insertion of further nucleic acids;

6. A vector comprising heterologous nucleic acid sequences in form of 2 or more cassettes each comprising at least one of different affinity tags, one consisting of one or more IgG binding domains of SPA, and at least one MCS linker for the insertion of further nucleic acids;

7. A cell containing a nucleic acid of claim 3 or a vector of claim 4; and

8. A reagent kit comprising a nucleic acid of claim 3 or a vector of claim 4, 5 or 6 for the expression of a fusion protein of claim 2 and support materials each capable of specifically binding one of the affinity tags.

USE - The methods can be used for the detection and or purification of substances capable of complexing with the fusion protein claimed. They can also be used for the detection and or purification of cells and or cell organelles expressing the fusion protein on their surface claimed. They can be used for studying the structure, activities or interactions with proteins or nucleic acids. The method not only facilitates efficient purification of protein of interest but also allows fishing for and detecting components present in complexes with which the polypeptides or subunits are associated or complexed either directly or indirectly, e.g. molecules such as linker mediators. This would allow selective fishing for certain substances which may be potential drugs, even from complex mixtures.

ADVANTAGE FOR USE IN DRUG
DEVELOPMENT APPLICATIONS

DETAILS - A method for detecting or purifying substances uses polypeptides or subunits fused to at least 2 different affinity tags at least one of which is an IgG binding domain of Staphylococcus aureus protein A (SPA).

[illegible] - A method for detecting or purifying substances uses polypeptides or subunits fused to at least 2 different affinity tags at least one of which is an IgG binding domain of Staphylococcus aureus protein A (SPA). [illegible]

a) providing an expression environment containing the one or more heterologous nucleic acids encoding at least 2 subunits of the protein complex, each being fused to at least one of different affinity tags, one of which consists of the one or more IgG binding domains of SPA;

b) maintaining the expression environment to express the polypeptides or subunits in a native form as fusion proteins with the affinity tags; and

c) detecting and/or purifying the complex by a combination of at least 2 different affinity purification steps each comprising binding the one or more subunits via the affinity tag to a support material capable of selectively binding one of the affinity tags and separating the complex from the support material after unbound substances have been removed.

INDEPENDENT CLAIMS are also included for the following:

1. a method for detecting and/or purifying a multiprotein and/or protein complexes comprising:

a) providing an expression environment containing the one or more heterologous nucleic acids encoding at least 2 subunits of the protein complex, each being fused to at least one of different affinity tags, one of which consists of the one or more IgG binding domains of SPA;

b) maintaining the expression environment to facilitate expression of the one or more subunits in a native form as fusion proteins with the affinity tags, and to allow formation of a complex between the one or more subunits and possibly other components capable of complexing with the one or more subunits; and

c) detecting and/or purifying the complex by a combination of at least 2 different affinity purification steps each comprising binding the one or more subunits via the affinity tag to a support material capable of selectively binding one of the affinity tags and separating the complex from the support material, after unbound substances have been removed;

2. fusion protein comprising at least one polypeptide or subunit of a protein complex fused to at least 2 different affinity tags, where one of the affinity tags consists of at least one IgG binding domain of SPA;

3. nucleic acid coding for a fusion protein of ...;

4. a vector comprising a nucleic acid as in 3 under the control of sequences facilitating the expression of a fusion protein thereof as in 2;

5. a vector comprising heterologous nucleic acid sequences in form of one or more cassettes each comprising at least 2 different affinity tags, one consisting of one or more IgG binding domains of Staphylococcus aureus protein A (SPA), and at least one PN linker for the insertion of further nucleic acids;

6. a vector comprising one or more nucleic acid sequences in form of one or more cassettes each comprising at least one of different affinity tags, one consisting of one or more IgG binding domains of SPA, and at least one PN linker for the insertion of further nucleic acids;

7. a cell containing a nucleic acid as in 3 or a vector of 4, 5 or 6; and

8. a reagent kit comprising a nucleic acid of 3 or a vector of 4, 5 or 6 for the expression of a fusion protein of 2 and support materials each capable of specifically binding one of the affinity tags.

USE - The method is for the detection and purification of multiprotein and/or protein complexes with low abundance in cells. The kit can be used in the detection and purification of multiprotein complexes from cells.

ADVANTAGE - The method has the advantage that it enables the detection and purification of multiprotein complexes with low abundance in cells.

facilitate efficient purification of polypeptides or subunits and/or complexes and/or their binding partners and/or to facilitate detection of polypeptides or subunits and/or complexes and/or their binding partners. Preferably, the affinity tags are not naturally occurring in the host cell, e.g. bacterial cell and/or have been modified so that they do not exhibit detectable activity, for certain substances which may be present in the complex mixtures.

NOVELTY - A method for detecting and/or purifying a polypeptide or subunit fused to at least 2 different affinity tags, one of which consists of one or more IgG binding domains of SPA.

DETAILED DESCRIPTION - A method for detecting and/or purifying a polypeptide or subunit of a protein, or protein complex comprising the polypeptide or subunits, e.g. complexes, multi-subunit complexes, cell organelles and cells, comprises:

a) providing an expression environment containing one or more heterologous nucleic acids encoding polypeptides and/or subunits of a biomolecular complex, the polypeptides or subunits being fused to at least 2 different affinity tags, one of which consists of one or more IgG binding domains of SPA;

b) maintaining the expression environment to express the polypeptides or subunits in a native form as fusions proteins with the affinity tags; and

c) detecting and/or purifying the polypeptide or subunits by a combination of at least 2 affinity purification steps, each comprising binding the polypeptides or subunits via the affinity tag to a support material capable of selectively binding one of the affinity tags and separating the polypeptides or subunits from the support material after unbound substances have been removed.

INDEPENDENT CLAIMS are also included for the following:

1) a method for detecting and/or purifying a biomolecular and/or protein complexes comprising:

a) providing an expression environment containing one or more heterologous nucleic acids encoding at least 2 subunits of a biomolecular complex, each being fused to at least one of different affinity tags, one of which consists of one or more IgG binding domains of SPA;

b) maintaining the expression environment to facilitate expression of the one or more subunits in a native form as fusions proteins with the affinity tags, and to allow formation of a complex between the one or more subunits and possibly other components capable of complexing with the one or more subunits; and

c) detecting and/or purifying the complex by a combination of at least 2 different affinity purification steps each comprising binding the one or more subunits via one affinity tag to a support material capable of selectively binding one of the affinity tags and separating the complex from the support material after unbound substances have been removed;

2) fusion proteins comprising at least one polypeptide or subunit of a protein complex fused to at least 2 different affinity tags, where one of the affinity tags consists of at least one IgG binding domain of SPA;

3) nucleic acid coding for a fusion protein of (2);

4) a vector comprising a nucleic acid as in (3) under the control of a sequence facilitating the expression of a fusion protein as in (2);

5) a vector comprising both 2 open nucleic acid sequences as in (3) of the multi-cistronic vector encoding at least 2 different affinity tags, one of which consists of one or more IgG binding domains of SPA, and at least one multiple cloning site for the insertion of a nucleic acid sequence;

6) [illegible]

[illegible] binding domain [illegible] DNA [illegible] for the [illegible] of further nucleic acids;

(7) a cell containing a nucleic acid of [illegible] or a vector of (4); and

(8) a reagent kit comprising a nucleic acid of [illegible] or a vector of (4) [illegible] for the expression of a fusion protein [illegible] and a [illegible] material capable of specifically binding [illegible] the affinity [illegible].

USE - The methods can be used for the detection and/or purification of substances capable of complexing with the fusion protein (claimed). They can also be used for the detection and/or purification of cells and/or cell organelles expressing the fusion protein on their surface (claimed). They can be used for studying the structure, activities or interactions with proteins or nucleic acids. The methods not only facilitate efficient purification of proteins of interest but also allows fishing for and detecting components present in complexes with which the polypeptides or subunits are associated or complexed either directly or indirectly, e.g. molecules such as linker mediators. This would allow selective fishing, for certain substances which may be potential drugs, even from complex mixtures.

WO 200119714A

CHOSEN DRAWING: Dwg.0/3

DERWENT CLASS: D16

CPI CODES: D05 H09; D05 H12E2; D05 H12E; D05 H14; D05 H17C1;